

WORLD INTELLECTUAL PROPERTY ORGANIZATION International Bureau



INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 7: WO 00/17221 (11) International Publication Number: C07H 21/04, C12Q 1/68 A1 (43) International Publication Date: 30 March 2000 (30.03.00) (81) Designated States: AU, CA, JP, US, European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, PCT/US99/19118 (21) International Application Number: (22) International Filing Date: 24 August 1999 (24.08.99) NL, PT, SE). (30) Priority Data: **Published** US With international search report. 24 September 1998 (24.09.98) 60/101,637 Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of (71) Applicant (for all designated States except US): DUKE UNI-VERSITY [US/US]; 230 North Building, Research Drive, P.O. Box 90083, Durham, NC 27712 (US). amendments. (72) Inventor; and (75) Inventor/Applicant (for US only): MEYER, Tobias [US/US]; 2628 McDowell Road, Durham, NC 27705 (US). (74) Agents: SIBLEY, Kenneth, D. et al.; Myers, Bigel, Sibley & Sajovec, P.A., P.O. Box 37428, Raleigh, NC 27627 (US).

(54) Title: METHOD OF MEASURING PROTEIN PROTEIN INTERACTIONS IN LIVING CELLS

(57) Abstract

A method of detecting protein-protein interactions in a living cell comprises (a) providing a cell that contains a first heterologous conjugate and a second heterologous conjugate, wherein the first heterologous conjugate comprises a first protein of interest conjugated to a detectable group, and wherein the second heterologous conjugate comprises a second protein of interest conjugated to a protein that expecifically binds to an internal structure within the cell, and then (b) detecting the presence or absence of binding of the detectable group to the internal structure, the presence of the binding indicating that the first and second proteins of interest specifically bind to one another. Additional aspects of the invention include nucleic acids encoding fusion proteins as described above, cells containing and expressing such fusion proteins, kits useful for carrying out the methods described above, and nucleic acid libraries useful as screening tools for carrying out the methods described above. The invention is useful for screening compounds for the ability to disrupt or inhibit the binding of known binding pairs and thereby identifying competitive inhibitors thereof. The invention is useful for screening one known protein of interest against a library of other proteins of interest to identify compounds that bind to the known protein of interest.